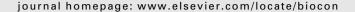# Potential of invasive and native solitary specialist bee pollinators to help restore the rare cowhorn orchid (*Cyrtopodium punctatum*) in Florida

Robert W. Pemberton[a], Hong Liu[b,*]

[a]Fairchild Tropical Botanic Garden, 2121 SW 28th Terrace Fort Lauderdale, FL 33312, USA
[b]University of Florida, Fort Lauderdale Research and Education Center, 3205 College Avenue, Fort Lauderdale, FL 33314, USA

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ABSTRACT

Cyrtopodium punctatum is a rare epiphytic orchid in southern Florida, made rare by historical over-collection. We examined the potential pollination of this orchid by the recently naturalized orchid bee (*Euglossa viridissima*), recorded as a pollinator of the orchid in tropical America, and found that this orchid bee is not a pollinator of the plant. We sought to learn what is responsible for relatively heavy fruit set in a Fairchild Tropical Botanic Garden population of *C. punctatum*, and determined that the native oil-collecting bee, *Centris errans*, is the most important pollinator. *C. punctatum* flowers at Fairchild have 18 times the fruit set of flowers in Everglades National Park. The difference is probably due to the many species and individuals of oil-reward plants in the Malpighiaceae in the garden, compared to one uncommon native species in the park. Female *C. errans* visit these oil-reward flowers to obtain edible oils to provision their brood. *Cyrtopodium* flowers appear to mimic the oil-reward flowers of the Malpighiaceae to attract *Centris* bee pollinators, much as many *Oncidium* orchids do in tropical America. We recommend that *Brysonima lucida*, a rare native malpighiaceous shrub, and *C. punctatum* be planted together in Everglades National Park and other natural areas to attempt to increase *C. errans* pollination, to restore and enhance the long-term survival of the orchid. Planting model malpighiaceous plants to enhance *Centris* bee pollination may be a useful restoration tactic for other rare orchid mimics in the American tropics, including Atlantic Forest in Brazil.

1. Introduction

Orchids are among the most popular ornamental plants. Their beauty, great diversity of form, fragrances, and exotic symbolism all contribute to their popularity. As of 2004, orchids were second after poinsettias in potted flowering plant sales in the United States with a wholesale value of $128 million, and orchid sales increased by five percent in 2004 (USDA, 2004). The great majority of orchids grown by gardeners and hobbyists originate from commercial mass production. Orchid collection from the wild, however, is an important problem despite CITES protection. But the collection of orchids from the wild is not a new phenomenon. Prior to the development of mass culture methods for orchids beginning in the 1940s, wild collected plants were the primary source for orchid cultivation (Arditti, 1992). These historical collections contributed to the

decline of many orchid species, which is exacerbated by continued illegal collection today (Seaton, 2007).

In this paper we deal with one such over-collected orchid, the once abundant but now rare cowhorn orchid, *Cyrtopodium punctatum* (L.) Lindl (see Fig. 1)., native to Florida and tropical America (Luer, 1972). There are interests and some efforts to restore this spectacular orchid in Florida, which is legally endangered in Florida (Coile and Garland, 2003). We studied *C. punctatum*'s pollination biology with the goal of assisting current and future restoration efforts. Knowledge of a rare plant's reproductive biology is essential for its conservation (Sipes and Tepedino, 1995).

van der Cingel (2001) indicated there is no data on the pollination of *C. punctatum* in Florida, or for other species in the genus *Cyrtopodium*. Dodson (1962) reported that a male orchid bee, *Euglossa viridissima* Friese, is a pollinator of *C. punctatum* in tropical America. *C. punctatum*, placed in the tribe Cymbidieae (Dressler, 1993), has recently been found to be basal in the Catasetinae, a tribe containing the perfume orchid genera *Catasetum, Cycnoches, Dressleria, Mormodes* (Chase et al., 2003), all of which have species visited by males of *E. viridissima* (Roubik and Hanson, 2004). This orchid bee has recently become naturalized in southeastern Florida (Skov and Wiley, 2005; Pemberton and Wheeler, 2006) and if it is able to pollinate *C. punctatum*, it could be important in the plant's restoration. The first objective of this study was to evaluate the role of *E. viridissima* as a pollinator of *C. punctatum*.

When sexual reproduction is absent or extremely low, as understood to be in *C. punctatum*, it can be exceptionally difficult to detect pollinators. We found a population of *C. punctatum* in Fairchild Tropical Botanic Garden (referred to as Fairchild hereafter) in Coral Gables, Miami-Dade County, Florida in which fruit production was occurring to a much greater degree than usually seen. Fairchild is within the historic range of *C. punctatum* and this population has been in the garden for a long time, with individual plants being known to senior gar-

den employees for more than 30 years (M. Collins, personal communication). This is considered a volunteer population because no accession records exist for these plants. The second objective of this study was to study the breeding system and pollination ecology of the heavily fruiting *C. punctatum* plants in Fairchild Garden.

2. Materials and methods

2.1. Study plant

C. punctatum is the most massive and one of the most distinctive orchids in southern Florida and the Neotropics. It has been known to science for almost 250 years, being one of the earliest tropical American orchids to be described (by Carl von Linne in 1759) (Bechtel et al., 1992). Also called the cigar orchid, due to its large fusiform pseudobulbs, the cowhorn orchid is usually epiphytic in Florida, but occasionally terrestrial in tropical America. Large plants can be one meter or more tall and bear paniculate, many flowered inflorescences. The 4–6 cm flowers are showy with greenish yellow sepals spotted with red–brown and bright yellow petals also spotted with red–brown. Wavy yellow–green floral bracts marked with red brown add to the color. The orchid is usually found in the forked branches of trees, or atop tree stumps, and can develop a large matted root system which serves as a platform supporting a bulky assemblage of pseudobulbs. The fruits are large pear-like capsules and take a full year to mature (Luer, 1972).

2.2. Breeding system and reward determination

To determine if the plant is autogamous, seven inflorescence tips with 26 flowers were bagged on March 31, 2007 on a large Fairchild plant, growing on the trunk of a palm (*Sabal domingensis* Becc.) from ca. 1.5 to 2.5 m above the ground. To determine if *C. punctatum* is self compatible, 12 flowers on two different potted plants at Fort Lauderdale were hand pollinated with self pollen. To determine outcross fruit set, 12 flowers from each of the same two potted plants were cross pollinated with pollen from the other plant between March 8 and 25, as sufficient numbers of flowers came into bloom. A paired-sample *T*-test was performed to determine the differences in fruit set between self and outcross treatments. At Fairchild, 14 flowers on the same single plant were treated with self pollen on March 23. This data was not included in the statistical analysis because no outcross treatments were done on the plant due to the inaccessibility of other *C. punctatum* plants, which grew high in palms and oaks. The flowers on the studied plants were also examined for the presence of nectar.

2.3. Potential pollination by E. viridissima

To examine the potential pollination of *C. punctatum* by *E. viridissima*, plants were studied in a Ft. Lauderdale, Broward County residential garden, where this bee was known to be common. Although Broward County is part of the plant's historic geographic range, no natural plants are currently known

Fig. 1 – Wagonload of native orchids, including the cowhorn orchid, *Cyrtopodium punctatum*, and the mule's ear orchid, *Trichocentrum undulatum*. The orchids were collected from Snake Hammock in what is now Everglades National Park and being taken to market in 1916 (photo and information: John Kunkle Small). The historical over-collection of *Cyrtopodium punctatum* made the orchid rare by the 1940s.

to occur there. We studied three cultivated plants believed to have originated in Florida and taken from the wild many years ago. The plants, which were in large baskets, were kept inside a screen house to exclude potential visitors, and then placed in a sunny place in the garden for the timed watches. Watches were conducted on nine days from March 4, 2007, when the flowers first began to open on the earliest flowering plant, to April 13, the end of the bloom on the latest flowering plant. During this period, several measures were made to assess the presence of the bee, its scent collecting and flower visiting behaviors. On March 5, a flowering perfume orchid (*Gongora powelli* Schltr.), known to attract male *E. viridissima* (Pemberton and Wheeler, 2006), was placed one and half meter away from an exposed *C. punctatum* for 10 min to detect male bees and see if they would visit the perfume orchid. On 12 March, a flowering plant of *Guarianthe skinneri* (Bateman) Dressler; W.E. Higgins, a food deception orchid pollinated by female *E. viridissima* (Pemberton, 2007, in press), was placed near the exposed *C. punctatum* plants for an hour during the watch. On March 15 a eugenol bait (eugenol placed on blotter paper) was placed within 2 m of flowering *C. punctatum* for 30 min to determine if male *E. viridissima* were present and interested to collect eugenol, a known attractant for this orchid bee (Roubik and Hanson, 2004). Observations for male *E. viridissima* were also made on an Africa basil (*Ocimum grattissimum* L.) growing 3 m from exposed *C. punctatum* plants. Previous observations indicated that male *E. viridissima* routinely collect oils from the leaves of this plant. All floral visitors to the exposed *C. punctatum* plants were observed and identified.

2.4. Timed watches for flower visitors at Fairchild Garden

Watches were made on the same large plant on which the breeding system determinations were made. Ideally, other plants should have been watched as well, but all other *C. punctatum* plants were growing high in oaks or at the tops of palms 8 m or more above the ground. Floral visitor observations, especially involving bees flying rapidly among hundreds of open flowers at a given time, was challenging even with the plant at eye level. Seeing and interpreting flower visitation in the masses of flowers 8–10 m high was not possible. Six watches were made on five different dates from March 16 to March 31, 2007, during which all flower visitors were observed and identified. During the March 16 watch, a eugenol bait was placed ca. 2 m of the observed plant to detect *E. viridissima*, which was not known to occur in the garden. The male success, i.e., pollinia removal rate, was obtained by non-destructive sampling of 4–7 inflorescences once a week, starting soon after the plant started to bloom until the end of the flowering season, for four weeks. Since individual flowers last ca. 13 days (personal observations), the pollinia removal rate over the entire season was estimated by summing the number of pollinia removed on the first and third weeks or the second and fourth weeks, depending on the inflorescence age, then dividing this sum by the total number of flowers sampled. On April 11 towards the end of the flowering season, three inflorescences from a different *C. punctatum* plant growing high in a different palm, *Attlalea butyracea* (Mutis ex L.f.) J.G.W. Borer, were clipped with pole pruner to assess pollinia removal and fruit formation at that

point in the season. The female success, i.e. the fruit set rate, of the primary study plant was determined at the end of flowering season.

2.5. Assessment of plant occurrence and reproduction

To determine the current presence of *C. punctatum* and reproductive status in southern Florida, we surveyed in Everglades National Park during a single day in October 2007, six months after flowering period, with the help of a park botanist who knew of the locations of some plants. Fruits present were derived from previous spring bloom. We sought information from a multi-year monitoring effort in Big Cypress National Wildlife Refuge, which with Everglades, is the place where the plant is known to be most abundant. We also surveyed the plants and their reproductive status at Fairchild in October 2007. The number of plants found, plants that had flowered this or last year as indicated by erect and drooping old inflorescences, and the number of fruit produced this year were counted. To estimate the fruit set rates (the number of fruit per flower per plant), we counted the number of flowers on 15 inflorescences on two plants in Fairchild during the flowering season. The total number of flowers produced by each plant was obtained by multiplying the number of inflorescences with the average number of flowers (47) per inflorescence. Fruit set rates were calculated by dividing the number of fruit per plant by the estimated number of flowers per plant. Binary logistic regression was used to determine if the number of flowers produced by each plant and the location (Everglades vs. Fairchild) were predictors of whether or not a flowering plant would produce at least one fruit. ANCOVA were used to determine whether or not plants in the two locations (Everglades vs. Fairchild) differed in the fruit rates (the number of fruit per flower per plant), using the number of flowers per plant as a covariate. Only fruiting plants were included in this analysis. We performed a similar ANCOVA on the number of fruits per plant (square root transformed).

3. Results

3.1. Breeding system and reward determination

None of the bagged inflorescence tips at Fairchild set fruit, indicating that *C. punctatum* is not autogamous and that a pollinator is needed to set fruit. The self pollination treatments of two cultivated plants resulted in 4/12 and 5/12 flowers setting fruit. The self pollination treatment of the large plant at Fairchild resulted in 2/14 flowers setting fruit. The outcross treatments with the potted plants resulted in 4/12 and 6/12 flowers setting fruit. This indicates that *C. punctatum* is self compatible and that there was no difference in fruit set between self and outcross pollination (Paired sampled T test, $t = -0.928$, df = 1, $P = 0.524$). This statistical result is based on two plants that were available for study; more plants would increase the confidence of the findings.

No floral nectar was found inside the flowers. Extrafloral nectar in the form of small but visible droplets was found on the flower buds, the lip and on the developing fruit. No macroscopic secretory glands were apparent with 10× magnification. A glucose test strip gave a positive test of the secre-

tion. A predatory vespid wasp (*Vespa dorsalis* Bequaert) frequently visited the secreting buds to collect the extrafloral nectar as did some halictid bees.

3.2. Potential pollination by Euglossa viridissima

The nine timed watches at Fort Lauderdale totaled 900 min or 15 h during which 473 flowers were watched in the three exposed plants. *E. viridissima* bees were present during all nine watches. Male bees collected the volatile oils from African basil leaves, visited flowers of the perfume orchid *G. powelli* and removed one pollinarium during the 10 min exposure of the orchid. Five male bees came to the eugenol bait during the 30 min baiting period. Male bees showed no interest in *C. punctatum* flowers. Female *E. viridissima* collected pollen by buzzing the flowers of *Solanum errianthum* D. Don, and collected nectar from *Pandorea jasminoides* (Lindl.) K. Schum. Female bees also visited flowers of *G. skinneri* and removed one pollinarium during the plant's one hour exposure period. All of these *E. viridissima* activities were within 1–4 m of the exposed *C. punctatum* plants. These bees appeared to be aware of *C. punctatum*. *E. viridissima* bees, probably females, were seen to inspect inflorescences on three occasions: one bee checked a few flowers by flying ca. 10 cm from them, another inspected some flower buds, and the third bee hovered before an inflorescence from about a 30 cm away. No *E. viridissima* was observed to touch or enter a *C. punctatum* flower.

3.3. Other flower visitors in Fort Lauderdale

Female *Centris nitida* Smith, an oil collecting bee (Apidae), regularly visited the *C. punctatum* flowers both as single and multiple bees, hovering before them and frequently touching the calli on the lips of the flowers. This bee, native to tropical America and only recently recognized as an exotic species in southern Florida (Pemberton and Liu, 2008), constituted 90% of the visitors to the Ft. Lauderdale plants. A *C. nitida* bee visited flowers and touched their lips 27 min after the first exposure of a plant, and 12 min after the sun shown on the exposed flowers on March 4. We counted the number of *C. nitida* bees and number of flowers visited during three watches and found that a total of 35 bees visited at least 151 flowers in 255 min. Despite *C. nitida*'s frequent visitation to *C. punctatum* flowers, only a single bee was observed (on March 25) to completely enter one flower, but it did not remove pollinia. After a watch on March 25, a female *C. nitida* was captured with a *C. punctatum* pollinarium attached to the back of its head. The viscidium was attached to the posterior surface of the head and the pollen sacks stood erect rising just above the dorsal surface of the head.

During the flowering period of *C. punctatum*, female *C. nitida* intensively collected the oil rewards from the flowers of *Brysonima lucida* (Mill.) DC. (Malpighiaceae) on a daily basis. This shrub, which is a native to Florida (Wunderlin and Hansen, 2003), grew about 15 m from the *C. punctatum* exposure area. During four different watches, four small green halictid bees (as yet unidentified) entered a total of seven flowers; three bees visited single flowers and one bee visited four. The bees completely entered the flowers but did not contact the columns or remove pollinia. An uncaptured small gray bee briefly entered two flowers on 12 March did not contact the column or remove pollinia. A few honey bees checked the flowers and flower buds but did not land. A single monarch butterfly (*Danaus plexipus* L.) probed a few flowers on one occasion but did not contact the column or remove pollinia.

Although no pollinia removal or deposition was observed, a total of four unlabeled fruit were found on two plants. This may have been due to natural, unobserved pollination, or the loss of tags from hand-pollinated flowers. Nine tags were missing from each of these plants. Most of these tags fell with aborted flowers to which they were probably attached. Limited natural pollination is suggested by the presence of a pollinarium found on an intact anther cap of a single flower, indicating probable pollen transport by a pollinator. This supplements the evidence provided by the captured *C. nitida* bearing a *C. punctatum* pollinarium.

3.4. Timed watches for flower visitors at Fairchild Garden

The five timed watches lasted a total of 635 min (ca. 10.5 h), and involved 526 flowers on a single continuously exposed plant. The watches began on March 16, when the observed *C. punctatum* had 147 flowers. The last watch was on March 31. *C. nitida* was present during three of the four watches. Nine bees were seen to visit 23 flowers but none were seen to fully enter flowers. Single or at times multiple bees touched the calli on the lips of many of the visited flowers but were not observed to enter any flowers. The most abundant and most meaningful visitor was female *Centris errans* Fox, another oil-collecting bee native to southeastern Florida (Pascarella, 2006). Female bees visited *C. punctatum* flowers during four of the five watches on all four dates. Eleven bees were observed to visit 41 flowers, and unlike *C. nitida*, were observed to completely enter at least 18 of these flowers. On March 16 and 31, *C. errans* were observed to remove pollinia from a single flower each day. The pollinia were attached to the back of their heads in the same position as the pollinarium attached to *C. nitida* captured in Ft. Lauderdale.

On March 15, female *C. errans* were observed to collect the oil reward from flowers of *B. lucida* plants, ca. 500 m from the *C. punctatum* plants. On May 8, after the watches ended, females of *C. errans* were observed to collect oils from the flowers of two non-native ornamental species belonging to the Malpighiaceae; the shrub *Galphimia gracilis* Bartl. and the vine *Stigmaphyllon sagraenum* A. Juss. The *S. sagraenum* grows on a rock wall ca. 20 m from the studied *C. punctatum*, while *G. gracilis* grew ca. 200 m away.

The other observed visitors were a honey bee which entered two flowers during one watch and a single leaf cutting bee *Megachile xylocopoides* Smith (Megachilidae), which touched the lips of five flowers during a single visit but entered none. The honey bee entered the flowers but did not remove pollinia. No *E. viridissima* came to the eugenol bait or was otherwise seen in the garden. The orchid bee is not known to yet occur in central or southern Miami-Dade County.

The studied *C. punctatum* plant was exposed to pollinators during its entire bloom. This allowed the determination of male and female success under more natural conditions.

The whole season pollinia removal rate (the number of pollinia removed per flower) was 0.317 (N = 526) and the fruiting rate (the number of fruit per flower) was 0.066. The late season assessment of the three inflorescences of a *C. punctatum* plant growing on *A. butyracea* found that they had 27, 21 and 24 flowers. These flowers had 20, 21 and 10 pollinia removed, respectively, which is an average one-time pollinia removal rate of 70.8%. The detectable fruit set in these inflorescences was 5, 8 and 4 fruit or an average of 23.6%.

3.5. Assessment of plant occurrence and reproduction at different sites

During the single day survey in Everglades, 16 plants were located. Other *C. punctatum* plants are known to park personnel, including at least 6 reproductive plants. In 2007, 10/16 located plants flowered, and 3/10 flowering plants produced fruit, one fruit each on three plants. In Fairchild, 6/12 plants flowered in 2007, and 3/6 flowering plants produced fruit, with the three fruiting plants producing 19, 13, and 9 fruit, respectively, a total of 41 fruit. Both the male and female reproductive components in the Fairchild plants were quite high with the midseason 70% pollinia removal and 13% fruit set in one sampled plant. The overall fruit set rate (fruits/flower) in Everglades was 0.00069 ± 0.0016 (mean ± std. dev.) compared to 0.0129 ± 0.0142 in Fairchild. The number of the flowers per plant was a marginally significant predictor of whether a plant would fruit and it was marginally significant (Wald = 3.182, df = 1, P = 0.074). Location (Everglades vs. Fairchild) was not a significant predictor of whether or not a plant would fruit (Wald = 0.709, df = 1, P = 0.400). Location, however, had a significant effect on the fruit rate in the ANCOVA (MS = 0.001, $F_{1,3}$ = 162.855, P = 0.001), with the Fairchild plants producing more fruit per flower. The effect of the number of flowers per plant on the rate of fruit set was not significant (MS = 0.00001, $F_{1,3}$ = 3.808, P = 0.146). The ANCOVA on the number of fruits per plant (square root transformed) produced similar results, which indicated that the number of fruit per plant at Fairchild was significantly larger than those at Everglades (MS = 9.358, $F_{1,3}$ = 33.195, P = 0.01). The effect of the number of flowers per plant on the number of fruit per plant was not significant (MS = 0.081, $F_{1,3}$ = 0.288, P = 0.629).

The *C. punctatum* survey in Big Cypress National Wildlife Refuge from 2002 to 2005 located 22 plants widely scattered throughout this large 729,000 acres preserve (J. Saddle personal communication). Ten of these were judged to be of reproductive size and during this four year period only two fruit were seen, one each on two different plants.

4. Discussion

4.1. Cyrtopodium punctatum pollination

The invasive orchid bee *E. viridissima* does not appear to be a pollinator of *C. punctatum*. Male *E. viridissima* were abundant during the *C. punctatum* exposures, and were readily attracted to fragrance sources related to their pollination activities, but they had no interest in the flowers of *C. punctatum*. Although *C. punctatum* flowers have fragrance, the fragrance is not the highly aromatic terpeniod fragrance of the perfume orchid mutualists of *E. viridissima*. Although female *E. viridissima* bees visit a wide array of plants for nectar, resin and pollen (Pemberton and Wheeler, 2006), the flowers of *C. punctatum* resemble (morphologically) none of those visited by the bees. These orchid bees can be deceived, as happens with afore mentioned *G. skinneri*. But in this nectar deceit orchid, the appearance of the lip of the flowers closely resembles the gullet type flowers of nectar reward flowers that the bees prefer (Pemberton, 2007, in press).

Our finding indicated that *C. errans* and *C. nitida*, to a lesser degree, were the only observed pollinators of this orchid in southeastern Florida. Subsequent to our study, we located a report of the pollination of *C. punctatum* by *C. errans* (Luer, 1972). Luer (1972) cited Adams (personal communication) to say that an oil collecting bee, *C. errans*, as *C. versicolor* (F.), is a pollinator of the orchid. We learned that this information was due to capture by Adams and Sauleda of two *C. errans* in Florida in 1970, with *C. punctatum* pollinia in a malaise aerial insect trap placed in the vicinity of *C. punctatum* (R. Sauleda personal communication). We located the captured specimens at Suzanne Koptur's laboratory at Florida International University in Miami and confirmed that the pollinia on the bees were from a *Cyrtopodium* species. *Cyrtopodium polyphyllum*, a naturalized Brazilian species was also present in Miami at the time (as *Cyrtopodium andersoni* R. Br., Luer, 1972).

On June 6, after the plants finished flowering, one female *C. errans* was collected at the Ft. Lauderdale garden site where *C. punctatum* were exposed. *C. errans* might have been present during the flowering of *C. punctatum* and possibly contributed to the limited natural fruit set found in the plants. The presence of *C. errans* in Broward County is beyond its reported geographical range (Pascarella, 2006), which suggests that this bee may occur more widely than understood. The *Centris* bee interactions with *C. punctatum* appear to represent a novel association involving oil reward members of the Malpighiaceae and rewardless *Cyrtopodium* orchid mimics. Females of *C. nitida* also pollinate *C. polyphyllum* (Vell.) Pabst ex F. Baros (Liu and Pemberton, submitted), a Brazilian orchid naturalized in southern Florida (Wunderlin and Hansen, 2003). This syndrome is the most common pollination system for *Oncidium* orchids. As in *Oncidium* flowers, female bees are focused on trying to obtain oil rewards which they perceive to be located on the callus of the lip or the interior of the flowers. Interestingly, *Oncidium* and *Cyrtopodium* are not closely related. *Cyrtopodium* is in the tribe Cymbidieae (Dressler, 1993) or the Catesetinae in the Maxillarieae (Chase et al., 2003), while *Oncidium* belongs to the Oncidinae in the Maxillarieae tribe (Dressler, 1993), indicating that the oil reward mimic pollination system has evolved more than once. Female *C. nitida* are pollinating ornamental *Oncidium sphacelatum*, native to Mexico and Central America, in Ft. Lauderdale (Pemberton, in press). *Cyrtopodium*, *Oncidium*, *Centris* bees and oil reward members of the Malpighiaceae are all limited to tropical and subtropical America (Mabberly, 1993; Michener, 2000) and this oil-deceit pollination syndrome probably functions widely in the region.

Our data suggest that *C. errans* is very likely to be the main pollinator of *C. punctatum* in Miami-Dade and Monroe counties. Everglades National Park lies within these counties and

Collier County. *C. errans* has been collected there, but *C. nitida* has not (Pascarella et al., 1999), but both could be present.

C. errans has a close association with *B. lucida* (Koptur, 2006), the native shrub belonging to the Malpighiaceae, the only wild native member of the family in southern Florida (Wunderlin and Hansen, 2003). This plant grows naturally in pine rockland and hammocks (tropical hardwood forest) in Miami-Dade County and Monroe County to the south and is rare (Wunderlin and Hansen, 2003). The upland habitats on which it naturally grows were mostly destroyed with the construction of Miami. Pine rockland now comprises only about 2% of the area that it previously occupied (Williams et al., 2007). We suspect that *C. errans* is now less common than it was historically because of the loss or decline of its principle food oil source—*B. lucida*. The bee's relative rarity may translate to the low fruit set observed in *C. punctatum* plants in Everglades.

C. nitida, which appears to be an occasional pollinator of *C. punctatum*, was first correctly identified in this study as naturalized bee native to Mexico to South America (Pemberton and Liu, 2008). However, neither of the *Centris* bees is known to occur in the northern Big Cypress National Wildlife Refuge or the Panther National Wildlife Refuge in Lee County, the northern part of the *C. punctatum* range (Pascarella, 2006; Pemberton and Liu, 2008). There are probably other as yet unknown pollinators of the orchid in these areas. Because *C. errans* is restricted to southern Florida and the Bahamas (Mitchell, 1962), *C. punctatum* must have different pollinators in the neotropical part of its range. As noted above these could be *Centris* species or oil collecting bees in related genera.

4.2. *Cyrtopodium punctatum reproductive success*

The mean fruit set at Fairchild plants was 18 times higher (1.29% vs. 0.069%) than in Everglades plants. It is not known if the orchid's low levels of fruit production in Everglades and Big Cypress has contributed to its population decline because it has not been demonstrated that the orchid population is seed limited. It is possible, however, that the wild population of *C. punctatum* has declined to such a degree that it is subjected to Allee effects, i.e. sub-optimal population performance associated with small populations (Allee et al., 1949). One mechanism for the Allee effects is through reduction of pollinator recruitment (Groom, 1998), which lead to a reduction in fruit set in non- autogamous plant species, such as *C. punctatum*. This may be particularly true in plants with specialized pollination systems (Kearns et al., 1998). We suspect that the presence of large *B. lucida* plants and other oil reward Malpighiaceae in Fairchild is responsible for this high fruit set in *C. punctatum*. The presence of a reward model has been shown to increase the fruit set in a rewardless orchid mimic (Anderson et al., 2005). We do not know what normal rates of fruit production were in *C. punctatum* in the wild prior to it and *B. lucida* being rare.

Deceit orchids typically have lower levels of pollinator visitation and fruit set than orchids with rewards (Neiland and Wilcock, 1998; Tremblay et al., 2005). Neiland and Wilcock list fruit set rates for 13 tropical food deceit orchids for which we calculated an average 7.5% fruit set, which is about the same as that observed in the Fairchild *C. punctatum*. Tremblay et al. (2005) reviewed the reproductive success of deceit and rewarding tropical orchids. Using this review, we calculated a 11.3% fruit set in deceit orchids, compared to 29.6% for reward orchids. Of the 27 species for which Tremblay et al. (2005) have pollinia removal rates, we calculated a removal rate of 42.1%, a bit higher than the average one-time removal rate of 30% that we measured for *C. punctatum* at Fairchild.

4.3. *Potential restoration of C. punctatum*

Our knowledge that *C. errans* is the primary pollinator of the orchid in Florida, instead of the orchid bee, can be used to help restore *C. punctatum*. Conservation efforts that promote pollinator visitation when fruit production is extremely low have been recommended by several authors (Kearns et al., 1998; Spira, 2001). A way to potentially boost the reproduction of the orchid is to increase its pollinator, *C. errans*, by planting *B. lucida* in or near the same sites with the orchid. The presence of the reward model for a rewardless deceit orchid can result in increased fruit set in the orchid (Anderson et al., 2005). The fruit set of a congeneric orchid, the invasive *C. polyphyllum* in southern Florida, was higher in residential gardens where more oil-reward plants were present than in a natural area where oil-reward plants was absent (Liu and Pemberton, submitted). The potential of planting *B. lucida* to promote *Centris* bees to increase pollination and fruit set in *C. punctatum* needs to be demonstrated, but the approach appears to be promising based our observations of higher numbers of *Centris* bees and pollination success in the orchid when malpighiaceous species are present or near. Everglades National Park is a good candidate for our proposed restoration method because it is developing native orchid restoration efforts, and has populations of *C. errans*, *B. lucida* and *C. punctatum* (Pascarella et al., 1999). It may be necessary to augment both populations of *C. punctatum* and *B. lucidia* for successful restoration of the orchid.

Planting model Malpighiaceae may help promote the reproduction in other rare oncidioid orchids with the *Centris*-oil reward plant (primarily Malpighiaceae) pollination syndrome in Florida and in other parts of tropical America. Oncidioid orchids typically have low fruit set, as with other deceit orchids, so may be particularly vulnerable when their numbers fall to low levels. Florida has three such orchids: *Trichocentrum undulatum* (SW.) Ackerman & M.W. Chase = *Oncidium undulatum* (Sw.) Salisb., *Tolumnia variegata* Braem (=*Oncidium bahamensis* Nash ex Brtton & Millsp) and *Oncidium ensatum* Lindl. (=*Oncidium floridanum* Ames). All three, which have small populations limited to small geographic areas, may be suitable candidates. In the Atlantic forest of Brazil some rare *Oncidium* species such as *O. hookeri* Rolfe, which hardly fruit (Alcantara et al., 2006), may also be suitable candidates for this restoration approach.

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REFERENCES

Alcantara, S., Semir, J., Solferini, V.N., 2006. Low genetic structure in an ephytic Orchidaceae (*Oncidium hookeri*) in the Atlantic rainforest of southeastern Brazil. Annals of Botany 98, 1207–1213.

Allee, W., Emerson, A., Park, O., Park, T., Schmit, K., 1949. Principles of Animal Ecology. W.B. Saunders Company, Philadelphia, Pennsylvania.

Anderson, B., Johnson, S.D., Carbutt, C., 2005. Exploitation of a specialized mutualism by a deceptive orchid. American Journal of Botany 92, 1342–1349.

Arditti, J., 1992. Fundamentals of Orchid Biology. John Wiley & Sons, New York.

Bechtel, H., Cribb, P., Launert, E., 1992. The Manual of Cultivated Orchid Species, third ed. The MIT Press, Cambridge, Massachusetts, USA.

Chase, M.W., Freeudenstein, J.V., Cameron, K.M., 2003. DNA data and Orchidaceae systematics: a new phylogenetic classification. In: Dixon, K.W., Kell, S.P., Barrett, R.L., Cribb, P.J. (Eds.), Orchid Conservation. Natural History Publications, Kota Kinabalu, Sabah, Malaysia, pp. 69–89.

Coile, N., Garland, M.A., 2003. Notes on Florida's endangered and rare plants. Botany Section No. 38, forth ed., Bureau of Entomology, Nematology and Plant Pathology, Division of Plant Industry, Florida Department of Agriculture and Consumer Services, Gainesville.

Dodson, C.H., 1962. The importance of pollination in the evolution of the orchids of tropical America. American Orchid Society Bulletin 31, 525–534.

Dressler, R.L., 1993. Phylogeny and Classification of the Orchid Family. Dioscoroides Press, Portland.

Groom, M.J., 1998. Allee effects limit population viability of an annual plant. American Naturalist 151, 487–496.

Kearns, C.A., Inouye, D.W., Waser, N.M., 1998. Endangered mutualisms: the conservation of plant-pollinator interactions. Annual Review of Ecology and Systematics 29, 83–112.

Koptur, S., 2006. The conservation of specialized and generalized pollination systems in subtropical ecosystems: a case study. In: Waser, N., Ollerton, J. (Eds.), Specialization and Generalization in Plant–Pollinator Interactions. University of Chicago Press. Chapter 15.

Luer, C.A., 1972. The Native Orchids of Florida. The New York Botanical Garden, New York.

Mabberly, D.L., 1993. The Plant Book. Cambridge, UK.

Michener, C.D., 2000. The Bees of the World. John Hopkins University Press, Baltimore, Maryland.

Mitchell, T. B., 1962. Bees of the eastern United States. Vol. 2. North Carolina Agricultural Experiment Station Technical Bulletin 152.

Neiland, M.R., Wilcock, C., 1998. Fruit set, nectar reward, and rarity in the Orchidaceae. American Journal of Botany 85, 167–1671.

Pascarella, J.B., 2006. Bees of Florida. <http://chiron.valdosta.edu/jbpascar/Intro.htm>. (Accessed November 2007).

Pascarella, J.B., Waddington, K.D., Neal, P.R., 1999. The bee fauna (Hymenoptera: Apoidea) of Everglades National Park, Florida and adjacent areas: distribution, phenology, and biogeography. Journal of the Kansas Entomological Society 72, 32–45.

Pemberton, R.W., 2007. Invasive orchid bee, *Euglossa viridissima*, pollinates the ornamental orchid (*Guarianthe skinneri*) in Florida. Lankesteriana 7, 461–468.

Pemberton, R.W., in press. Pollination of the ornamental *Oncidium sphacelatum* by the naturalized oil-collecting bee (*Centris nitida*) in Florida. Selbyana.

Pemberton, R.W., Liu, H., 2008. The naturalization of an oil collecting bee *Centris nitida* in Florida and the eastern United States, with notes on the *Centris* species native to Florida. Florida Entomologist 91, 101–109.

Pemberton, R.W., Liu, H., in press. Naturalization of *Dalechampia scandens* in southern Florida. Caribbean Journal of Science.

Pemberton, R.W., Wheeler, G.S., 2006. Orchid bees don't need orchids: evidence from the naturalization of an orchid bee in Florida. Ecology 87, 1995–2001.

Roubik, D.W., Hanson, P.E., 2004. Orchid Bees of Tropical America, Biology and Field Guide. Instituto Nacional de Biodiversidad, San Jose, Costa Rica.

Seaton, P.T., 2007. Orchid conservation: where do we go from here? In: Pupulin, F., (Ed.), Proceedings of 3rd International Orchid Conservation Congress/2nd International Conference on Neotropical Orchidology. Lankesteriana 7, pp. 13–16.

Sipes, S.D., Tepedino, V.J., 1995. Reproductive biology of the rare orchid *Sprianthes diluvialis*: breeding system, pollination, and implications of conservation. Conservation Biology 9, 929–938.

Skov, C., Wiley, J., 2005. Establishment of the Neotropical orchid bee *Euglossa viridissima* (Hymenoptera: Apidae) in Florida. Florida Entomologist 88, 225–227.

Spira, T.P., 2001. Plant–pollinator interactions: a threatened mutualism with implications for the ecology and management of rare plants. Natural Areas Journal 21, 78–88.

Tremblay, R.L., Ackerman, J.D., Zimmerman, J.K., Calvo, R.N., 2005. Variation in sexual reproduction in orchids and its evolutionary consequences: a spasmodic journey to diversification. Biological Journal of the Linnean Society 84, 1–54.

USDA National Agricultural Statistics Survey, 2004. Floriculture Crops Survey.

van der Cingel, N.A., 2001. An Atlas of Orchid Pollination: America, Africa, and Australia. A. A. Balkema, Rotterdam, Netherlands.

Williams, D.A., Wang, D.A., Borchetta, M., Gaines, M.S., 2007. Genetic diversity and spatial structure of a keystone species in fragmented pine rockland habitat. Biological Conservation 138, 256–268.

Wunderlin, R.P., Hansen, B.F., 2003. Guide to the Vascular Plants of Florida, second ed. University Press Florida, Gainesville.